SOME EXPERIMENTS ON LARVICIDES

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At present the substance in most general use as a larvicide is petroleum, which acts by forming a thin film on the surface of the water, thereby drowning the larvae when they rise to the surface to breathe. In the case of enclosed bodies of water, such as wells and tanks, this usually acts admirably at first, but in the course of a short time the film, unless very thick, gets broken. This happens much more readily in pools or stagnant places on river banks where mosquitos breed. The film of petroleum in these cases soon gets broken or blown to one side sufficiently to allow the mosquitos to lay their eggs, and the larvae to develop into mosquitos. Moreover, the presence of grass or weeds on the banks of these pools prevents the formation of an unbroken film over the whole surface, and this permits the reproductive processes to go on more or less uninterruptedly.

Petroleum has other disadvantages arising from its inflammability and its liquid nature, which render it less convenient for transit.

An ideal larvicide would appear to be a solid substance which kills larvae even when used in the form of an extremely dilute solution. This would be convenient for use, as no great weight of material would have to be carried from place to place. The solution, when diluted to the maximum point at which it will still kill larvae in a reasonable time must also be harmless to men and domestic animals if the water is liable to be used for drinking purposes. Should there be no chance of the water being drunk, however, a stronger solution may be used, and the larvae killed more quickly.

We have tried a number of solutions as larvicides, and an account of the results obtained with those substances which we thought might be most suitable is given in the following pages. Most of our experiments were carried out with the larvae of *Culex pipiens*, about fifty larvae being used in each case.

We first tried the effect of a larvicide recommended by Le Prince, quoted by Ross (1910), prepared by dissolving resin in crude carbolic acid, and treating the solution with caustic soda. We found it difficult to obtain a proper emulsion without sediment, however, as the resin did not dissolve readily in the carbolic acid. This larvicide has been used with success in America, where it is said to cost only sevenpence a gallon, the average mixture containing 300 gallons crude carbolic acid, 200 pounds resin, 30 pounds caustic soda.

Our results with this larvicide are as follows:—

One part larvicide in 500 parts of water killed 80 per cent. of the larvae (*Culex pipiens*) in two hours, and the remainder in three and a half hours.

One part larvicide in 1,000 parts of water killed 70 per cent. of the larvae in four hours, and most of the remainder by next morning (eighteen hours). One larva, however, lived in this solution for twenty-two hours.

One part larvicide in 2,000 parts of water killed about 30 per cent. of the larvae in six hours, but the rest of the larvae were killed very gradually, some surviving for about fifty hours.

We used the different ingredients in the proportions mentioned above in the preparation of this larvicide, the value of which seems to depend very largely upon the composition of the crude carbolic acid used. Le Prince found this larvicide to act more quickly and at a greater dilution than we did, perhaps because he employed a stronger carbolic acid from which to make the final product.

We next tried an emulsion prepared by the 'Sanitas' Company, Limited, called 'Sanitas-Okol.' This appears to contain a large proportion of phenols and allied compounds, and when much diluted mixes well with water.

We obtained the following results with 'Sanitas-Okol,' using *Culex pipiens* larvae.

One part 'Sanitas-Okol' to 600 parts of water killed all the larvae in fifteen minutes.

One part 'Sanitas-Okol' to 1,000 parts of water killed about 40 per cent. of the larvae in fifteen minutes, and the rest in seventy-five minutes. Many of the pupae were nearly ready to hatch out into mosquitos, at which stage we frequently found them to be more resistant to the action of larvicides, perhaps because they are then less firmly attached to the outer shell, which prevents the solution reaching their tissues so readily.

One part 'Sanitas-Okol' in 2,000 parts of water killed 50 per cent. of a young lot of larvae in sixteen minutes, and the rest in twenty-eight minutes.

One part 'Sanitas-Okol' in 2,500 parts of water killed all the larvae except one in seventy minutes, the last being dead in ninety minutes.

One part of 'Sanitas-Okol' in 5,000 of water killed 40 per cent. of the larvae in seventy minutes, and the rest in two and a half hours.

In another experiment this dilution (one in 5,000) killed 50 per cent. of the larvae in one and three-quarter hours, and all but three in three and a half hours, the last three being dead in about five hours.

One part 'Sanitas-Okol' in 10,000 parts of water killed all the larvae except two in six hours, the last two dying in the course of the night.

With one part 'Sanitas-Okol' in 20,000 parts of water about 10 per cent. of the larvae lived for twenty-one hours, while two larvae were still alive after another twenty-four hours.

We have also tried the effect of 'Sanitas-Okol' on the larvae of *Anopheles bifurcatus*, and found as follows:—

One part 'Sanitas-Okol' to 5,000 parts of water killed eighteen larvae out of twenty in two and a quarter hours, the other two living for three and a quarter hours.

One part 'Sanitas-Okol' in 10,000 parts of water killed twenty-three larvae out of twenty-six in five and a half hours, and the rest in about eight and a half hours.

It will be seen that 'Sanitas-Okol' acts very powerfully as a larvicide, and can be used quite satisfactorily in dilutions up to 1 in 10,000. It is also quite non-poisonous at this great dilution.

In view of the fact that mercuric chloride acts so well as a germicide, we thought it well to try the effect of solutions of this compound on Culex larvae.

One part mercuric chloride in 2,000 parts of water only killed the larvae extremely slowly, one larva living for three days.

One part mercuric chloride in 1,000 parts of water killed all the larvae except one in eighteen hours.

We carried out no further experiments with mercuric chloride, as it is evidently unsuitable as a larvicide from its poisonous nature and comparatively feeble action on larvae. It would also be too expensive to use on a large scale.

The same objections can be taken to the use of copper sulphate, which failed to kill all the larvae in two days, even when used in the form of a solution containing 1 part of copper sulphate to 500 parts of water. A solution of cupric hydrate in ammonia, containing the same proportion of copper as mentioned above, had no better effect as a larvicide, although one part of ammonia solution (34 per cent.) to 125 parts of water killed all the larvae in twenty minutes.

Oxalic acid was not found to act powerfully on Culex larvae. A solution containing one part of the acid in 1,000 parts of water killed the larvae slowly, one surviving eighteen hours, and when one part of oxalic acid in 2,000 parts of water was used 10 per cent. of the larvae lived for about thirty hours.

It may be of interest to add that saponin (one part to 400 parts of water) was found to have no effect on larvae. While we had scarcely considered it in the light of a possible larvicide, we thought it worth while to try the effect of keeping larvae in such a solution, in view of the haemolytic action of saponin on the blood of higher animals.

We have finally turned our attention to potassium cyanide as a larvicide. As this substance is very much more poisonous than any others we had employed we did not use solutions as dilute as one in 1,000, which would be unsafe should there be any possibility of any water thus treated being drunk.

The following results were obtained with potassium cyanide, using *Culex pipiens*.

One part cyanide in 26,000 of water killed all the larvae in less than two hours.

One part cyanide in 58,000 of water killed all the larvae in three and one-third hours.

One part cyanide in 106,000 parts of water killed all the larvae in five hours.

In another experiment with nearly the same strength (one in 110,000), all the larvae except two were killed in five hours, the last two living about seven and a half hours.

One part cyanide in 240,000 parts of water killed all the larvae in the course of a night (less than sixteen hours from the time the cyanide was added).

One part cyanide in 303,000 parts of water killed about 50 per cent. of the larvae in six and a half hours, and all except one in twenty-two hours.

We have not carried out any experiments with weaker solutions of potassium cyanide than last mentioned, as we did not consider it advisable to use solutions in which larvae can live for a longer period than about eighteen hours, and a solution of one part cyanide to 300,000 parts of water was found by repeated experiment always to satisfy this condition, though in some cases individual larvae survived about twenty-four hours.

We have tried the effect of a solution containing one part potassium cyanide to 300,000 parts of water on two lots of Anopheles larvae, and in both cases about 80 per cent. of the larvae were killed in less than eight hours, and the rest in from twelve to fifteen hours.

Strychnine in dilutions greater than one in 50,000 parts of water appeared to have no effect on larvae. As we had obtained much more satisfactory results with potassium cyanide we did not try solutions of strychnine stronger than the above.

We have been unable to discover any other substance whose potency as a larvicide approaches that of potassium cyanide. This compound also possesses the advantage of being easily carried about and of being comparatively cheap. Its one drawback lies in the fact that it is extremely poisonous, and it is apt to undergo partial hydrolysis with the production of a certain proportion of free prussic acid.

On the other hand the use of potassium cyanide can be restricted to stagnant water which is not used for drinking purposes, while it must be remembered that water containing cyanide in the proportion of one to 250,000 or 300,000 would have to be consumed in very large quantities before having any deleterious effects.

We have endeavoured to find the most convenient form in which to use potassium cyanide as a larvicide. It is, of course, most conveniently carried about in the solid form, but if a piece be simply thrown into a pool of water it sinks, and although it is very readily dissolved the solution may not diffuse rapidly through the whole pool. Accordingly, we tried mixing the cyanide with a floating soap, both in the form of powder, and compressing the mixture into pills or tablets. These will still float, provided that the proportion of cyanide to soap is not too high, and that too great pressure is not used in the preparation of the tablets. In the latter case the resulting tablets are too dense, owing to there being but little air retained in the substance, and consequently they sink. The advantage of using these floating tablets is that the cyanide soon dissolves out, and the solution being heavier than water diffuses comparatively rapidly through the whole mass of the water, and thus practically none of the larvae escape exposure to the action of the drug. Other substances of a light and porous nature may, of course, be used instead of soap as a medium for the preparation of floating tablets containing the requisite amount of cyanide. These substances should preferably be insoluble, or at least of a comparatively inert nature.

A convenient size of tablet is one containing three or four grains of cyanide, which will suffice for twelve or sixteen gallons of water. These should be sealed up in tins of about 100 tablets, and, of course, all precautions must be taken to prevent any danger of tablets being eaten by children or others. Instructions ought also to be given that no water which is to be used for drinking purposes is to be treated with cyanide. With these precautions there is practically no danger of any harm resulting from the use of potassium cyanide in this form as a larvicide. It is unnecessary to refer here to previous experiments on larvicides, as these are familiar to all students of the subject.

REFERENCE

Ross, Sir R. Prevention of Malaria (John Murray), section 43.